

CLAIMS

Claim 1-11 (canceled)

12. (currently amended) A method for generating a new catalytic activity in an enzyme, comprising the steps of:

- a) introducing a DNA sequence coding for the enzyme into the *Escherichia coli* strain XL1-Red or into a functional derivative thereof which is also an *E. coli* strain carrying carries the genetic markers *relA1*, *mutS*, *mutT* and *mutD5* and having an increased mutation rate,
 - b) incubating the transformed *Escherichia coli* strain XL1-Red or its functional derivative to generate mutations in the DNA sequence,
 - c) transferring the mutated DNA sequence from the transformed *Escherichia coli* strain XL1-Red or its functional derivative to a microorganism which has no impeding enzyme activity which would impede selection,
 - d) incubating this microorganism to detect the new catalytic enzyme activity in at least one selection medium which comprises at least one enzyme substrate to recognize the newly generated catalytic activity in the enzyme, with or without other indicator substances, and
 - e) selecting the microorganisms which show the newly generated catalytic activity, said microorganisms in steps c), d) and e) being a member selected from the group consisting of bacteria, fungi and yeasts,
- wherein the enzyme is selected from the group consisting of lipases, amidases, nitrilases, ether hydrolases, peroxidases, glycosidases and phytases.

13. (previously presented) The method of claim 12, wherein the enzyme is a lipase.
14. (previously presented) The method of claim 12, wherein the enzyme is an amidase.
15. (previously presented) The method of claim 12, wherein the enzyme is a nitrilase.
16. (previously presented) The method of claim 12, wherein the enzyme is an ether hydrolase.
17. (previously presented) The method of claim 12, wherein the enzyme is a peroxidase.
18. (previously presented) The method of claim 12, wherein the enzyme is a glycosidase.
19. (previously presented) The method of claim 12, wherein the enzyme is a phytase.
20. (previously presented) The method of claim 13, wherein the lipase is selected from the group of lipases consisting of *Pseudomonas cepacia* lipase PS, *Pseudomonas cepacia* lipase AH, acylase, *Rhizopus delamar* lipase, *Rhizopus javanicus* lipase, *Candida rugosa* lipase, *Mucor javanicus* lipase, *Penicillium roquefortii* lipase, *Penicillium cyclopium* lipase, *Chromobacterium viscosum*

lipase, *Rhizomucor miehei* lipase, *Humicola lanuginosa* lipase, *Candida antarctica* lipase B and *Candida antarctica* lipase A.

21. (previously presented) The method of claim 12, wherein steps (a) to (e) are performed several times in sequence by reisolating and retransforming the DNA sequence from the microorganisms selected in step (e) to the strain *Escherichia coli* XL-1 Red or its functional derivative.
22. (currently amended) A method for generating a new catalytic activity in an enzyme, comprising the steps of:
- a) introducing a DNA sequence coding for the enzyme into the *Escherichia coli* strain XL1-Red or into a functional derivative thereof which is also an *E. coli* strain carrying carries the genetic markers *relA1*, *mutS*, *mutT* and *mutD5* and having an increased mutation rate,
 - b) incubating the transformed *Escherichia coli* strain XL1-Red or its functional derivative to generate mutations in the DNA sequence,
 - c) transferring the mutated DNA sequence from the transformed *Escherichia coli* strain XL1-Red or its functional derivative to a microorganism which has no impeding enzyme activity which would impede selection,
 - d) incubating this microorganism to detect the new catalytic enzyme activity in at least one selection medium which comprises at least one enzyme substrate to recognize the newly generated catalytic activity in the

enzyme, with or without other indicator substances, and
e) selecting the microorganisms which show the newly generated catalytic activity, said microorganisms in steps c), d) and e) being a member selected from the group consisting of bacteria, fungi and yeasts, wherein the enzyme is an esterase selected from the group consisting of *Pseudomonas fluorescens* esterase, pig liver esterase and *Thermoanaerobium brockii* esterase.

23. (previously presented) The method of claim 22, wherein steps (a) to (e) are performed several times in sequence by reisolating and retransforming the DNA sequence from the microorganisms selected in step (e) to the strain *Escherichia coli* XL-1 Red or its functional derivative.

24. (new) A method for generating a new catalytic activity in an enzyme, wherein the new catalytic activity is within the same International Union of Biochemistry class as the enzyme's original catalytic activity, comprising the steps of:

- a) introducing a DNA sequence coding for the enzyme into the *Escherichia coli* strain XL1-Red, or into a functional derivative thereof which is also an *E. coli* strain carrying the genetic markers *relA1*, *mutS*, *mutT*, and *mutD5*, and having an increased mutation rate,
- b) incubating the transformed *Escherichia coli* strain XL1-Red or its functional derivative to generate mutations in the DNA sequence,

- c) transferring the mutated DNA sequence from the transformed *Escherichia coli* strain XL1-Red or its functional derivative to a microorganism which has no enzyme activity which would impede selection,
- d) incubating this microorganism to detect the new catalytic activity in at least one selection medium which comprises at least one enzyme substrate to recognize the newly generated catalytic activity, with or without other indicator substances, and
- e) selecting the microorganisms which show the newly generated catalytic activity, said microorganisms in steps c), d) and e) being a member selected from the group consisting of bacteria, fungi and yeasts, wherein the enzyme is selected from the group consisting of lipases, amidases, nitrilases, ether hydrolases, peroxidases, glycosidases, phytases, and esterases selected from the group consisting of *Pseudomonas fluorescens* esterase, pig liver esterase and *Thermoanaerobium brockii* esterase.

25. (new) The method of claim 24, wherein the enzyme is a lipase.

26. (new) The method of claim 25, wherein the lipase is selected from the group of lipases consisting of *Pseudomonas cepacia* lipase PS, *Pseudomonas cepacia* lipase AH, acylase, *Rhizopus delamar* lipase, *Rhizopus javanicus* lipase, *Candida rugosa* lipase, *Mucor javanicus* lipase, *Penicillium roquefortii* lipase,

Penicillium cyclopium lipase, *Chromobacterium viscosum* lipase, *Rhizomucor miehei* lipase, *Humicola lanuginosa* lipase, *Candida antarctica* lipase B and *Candida antarctica* lipase A.

27. (new) The method of claim 24, wherein steps (a) to (e) are performed several times in sequence by reisolating and retransforming the DNA sequence from the microorganisms selected in step (e) to the strain *Escherichia coli* XL-1 Red or its functional derivative.

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